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(54) Title: REDUCTION OF ACRYLAMIDE FORMATION IN FOOD PROCESSING

(57) Abstract: The invention relates to the use of acid treatment of carbohydrate-containing foodstuffs prior to cooking by roasting, frying, grilling or baking whereby to reduce the acrylamide content of the cooked product.

Reduction of acrylamide formation in food processing

The present invention relates to improvements in and relating to cooked food, in particular vegetables which are fried, grilled, baked or roasted.

In a publication by the Swedish National Food Administration (see www.slv.se/engdefault.asp) it was reported that many cooked foods, in particular fried, grilled or baked foods, had surprisingly been found to contain high levels of the toxic contaminant acrylamide. No suggestion was made as to how the acrylamide context of such foods could be reduced.

A further report of acrylamide production in food cooking occurred in Tareke et al., J. Agric. Food Chem 50: 4998-5006 (2002).

We have now surprisingly found that the acrylamide content of cooked foods can be reduced by treatment of the food prior to cooking with lactic acid generating microorganisms and/or with acid.

Thus viewed from one aspect the invention provides the use of a lactic acid producing microorganism for the treatment of a food material to reduce acrylamide production in subsequent cooking thereof.

Lactic acid producing microorganisms are well known and examples include lactic acid bacteria such as Bifidobacterium sp., Brevibacterium sp., Lactobacillus sp., Lactococcus sp., Leuconostoc sp., Micrococcus sp., Oenococcus sp., Pediococcus sp., and Streptococcus sp. Lactobacilli are especially preferred for use according to the invention, in particular Lactobacillus plantarum strains NCDO 1752 and NCDO 1193 (available from the National Collection of Food Bacteria) and Lactobacillus NCIMB 40450. Other strains of lactobacillus which generate lactic acid and are safe for use in foodstuff treatment have been described widely in the scientific literature.

The treatment with a lactic acid producing microorganism according to the invention preferably involves incubation in an aqueous medium for up to 7 days, e.g. 30 minutes to 24 hours, especially 1 to 6 hours. Incubation is preferably at 4 to 45°C, e.g. 25 to 35°C, i.e. as is conventional for such microorganisms.

Typically such treatment may involve homofermentative lactic acid bacteria incubation in an aqueous medium.

Viewed from a further aspect the invention provides the use of a physiologically acceptable acid for the treatment of a food material to reduce acrylamide production in subsequent cooking thereof.

The physiologically tolerable acid used according to the invention may be any acid acceptable for use in foodstuffs, e.g. organic acids, such as citric, malic, acetic, maleic, tartaric, succinic and lactic acids or inorganic acids such as hydrochloric, sulphuric and phosphoric acids and sulphur dioxide. The use of citric and hydrochloric acids is especially preferred, as is the use of lactic acid and/or of phosphoric acid. For cereal or potato-based foodstuffs, the use of hydrochloric acid is especially preferred while for cereal-based products the use of lactic acid is particularly preferred. The acid is preferably used in a quantity and strength sufficient to reduce the surface pH of the food material treated to 1 to 5.5, preferably 3 to 5, especially about 4. Following acid treatment, the food material is preferably stored for up to 7 days (e.g. 30 minutes to 24 hours, especially 1 to 6 hours before cooking or freezing.

In this process, the acid is preferably used in the form of a buffer solution.

Following treatment with the acid and/or the lactic acid producing microorganism, the food material may be cooked using cooking techniques that expose the material

to temperatures above 150°C, e.g. by baking, grilling, roasting or frying.

Before such high temperature cooking, it is desirable to rinse the treated food material with water.

The cooking may be a single stage operation. However it may instead be one stage of a multi stage (e.g. two stage) cooking procedure. Thus the technique of the invention is especially applicable to food materials which are treated according to the invention, partially cooked, transported and/or stored, then cooked again.

The food material treated according to the invention may be any carbohydrate-containing food material but especially preferably is a plant or plant-derived material, e.g. a vegetable or cereal, in particular a root vegetable or a tuber (e.g. potato). Especially preferably the food material is potato, yam, onion, carrot, swede, turnip or parsnip. Such food materials are preferably processed (e.g. peeled, diced, sliced, chipped or chopped) prior to treatment according to the invention.

The invention is also particularly applicable to products made from vegetable (e.g. potato) or cereal (e.g. rice, barley, wheat, rye, oat, maize, etc.) flours, granulates or fragments, in particular breads (especially crisp-breads, biscuits, wafers, cookies and crackers), cakes, snacks (e.g. crisps (in American-English chips), pretzels, and the like), breakfast cereals (e.g. "cornflakes" and the like), restructured french fries, potato-croquettes, and to granulated and coffees and cocoas.

Thus in a further aspect the invention provides a process for the production of a food product which comprises fermenting a granulated or crushed carbohydrate-containing plant material with a lactic acid producing microorganism, optionally formulating the fermented material into a shaped product (e.g. by

extrusion, rolling or moulding a paste or dough), and cooking to produce said food product.

In place of fermentation, acid treatment as described above may be used: however this is less preferable.

The granulated carbohydrate-containing plant material, e.g. a potato or cereal flour, may be mixed with un-treated granulated carbohydrate-containing plant material before cooking. Desirably the treated:untreated weight ratio is from 25:75 to 100:0, especially 50:50 to 95:5.

The term granulate or granulated as used herein may where the context permits include fine to coarse particulates, e.g. flours, granules, grits, fragments, etc. Preferably however the granulates will be 2mm or smaller in maximum dimension.

In the case where the plant material is coffee bean, granulation, fragmentation or crushing may be omitted: however this is also less preferable. Moreover the cooked food product in this instance may be further processed, e.g. by conventional means, to produce dried instant coffees or liquid coffee extracts.

The food products thus produced are desirably packed into sealed, preferably sterilized containers, e.g. cartons, plastic or foil bags, bottles, etc. Particularly preferably such containers are moisture-proof.

Where appropriate, the food products produced according to the invention may be further processed, e.g. by drying, freezing, cutting, decorating, etc. Such processing steps, which are often conventional for the particular food product, form further optional steps in the processes of the invention.

However the invention is especially applicable for the production of french-fried potatoes, in particular so-called oven-ready french-fried potatoes which are provided to the consumer in part-cooked form for baking

prior to serving, as well as to the production of chopped ready-to-fry potatoes (e.g. of the type produced for deep frying in restaurants).

Thus viewed from a further aspect the invention provides a process for the preparation of ready to cook (e.g. oven-ready or ready to fry) french fried potatoes which process comprises chopping potatoes, fermenting the chopped potatoes with a lactic acid producing microorganism, frying the fermented chopped potatoes, and optionally loading the fried fermented chopped potatoes in a container, and optionally sealing the container.

Viewed from a still further aspect the invention provides a process for the preparation of ready to cook (e.g. oven-ready or ready to fry) french fried potatoes which process comprises chopping potatoes, treating the chopped potatoes with a physiologically acceptable acid, frying the acid treated potatoes, and optionally loading the fried potatoes into a container, and optionally sealing the container.

The chopping stage in this process is preferably such as to produce batons having a cross-sectional area of 10 to 100mm^2, especially preferably 25 to 80mm^2.

This process can also be used for the preparation of hash browns.

The container used in these processes will typically be a plastic bag, paper carton or bag or other container conventionally used for storage and transport of ready to cook french fries.

Viewed from a still further aspect the invention thus also provides a container containing ready to cook (e.g. oven-ready or ready to fry) french fried potatoes produced by frying chopped potatoes pre treated with a lactic acid producing microorganism and/or with a physiologically acceptable acid.

The potatoes treated according to the invention are preferably of a variety selected from Maris Piper, Beate

or Russet, especially Maris Piper. Saturna, King Edward, Russet Burbank, Bintje, Shepady and Shasta may also be used. Especially preferably the potatoes are selected from varieties having a reducing sugar content of less than 1.5% wt, particularly less than 1.0% wt.

French-fries are preferably made from sliced potato; however they may also be made from extruded or moulded carbohydrate-containing pastes produced using powdered or granulated potato and/or cereal (e.g. rice).

Besides being useful in the production of french-fried potatoes, the invention is also especially applicable in the production of potato crisps (also known in America as potato chips). In this regard, the acid and/or microorganism treatment is preferably effected on the sliced potato prior to frying.

The invention is also applicable to grain, i.e. cereal, products, e.g. breads, biscuits (known in America as cookies), and in particular crisp breads. In this aspect of the invention, the treatment according to the invention may be effected using lactic acid bacteria in the production of the dough and/or by acid treatment (e.g. treatment with sulphur dioxide or hydrogen chloride) of the flour.

Besides the fermentation and/or acid treatment according to the invention, the food products of the invention may be prepared by conventional methods, optionally involving rinsing and/or drying after the treatment. Thus such food products may optionally contain further components, such as conventional foodstuff components or additives, e.g. salt, sugars, flavours, fruit, fruit extracts, nuts, eggs, milk, flour, bread, breadcrumbs, stabilizers, colours, buffers, acidulants, yeast, bicarbonate, etc.

The invention will now be illustrated further with reference to the following non-limiting Examples.

Example 1

Crisp bread

Ingredients

Finely ground whole rye flour was obtained from Cerealia Mills, Oslo, Norway.

Crisp bread recipe

The crisp bread dough was based on a standard recipe made from 1150g rye flour, 1000g water, 20g NaCl. The ingredients were mixed and the dough was rolled out to proper thickness and baked in a stone oven at 240°C.

Acid equivalent

Total amount of acid in the flour-water mixtures was determined as Acid equivalents, S°, by titration. To 10g flour-water mixture, 90g of distilled water is added. During stirring, 0.1 N NaOH is added until a stable pH of 8.5 is reached. Acid equivalents is expressed as the amount of 0.1 N NaOH consumed, in ml.

Pre-treatments

Fermentation

The rye flour used in the crisp bread dough was fermented using a lactic acid bacteria according to the following description: 1000g rye flour and 1000g tap water at 30°C was mixed and bacteria (10^6 bacteria/g flour) was added. The bacteria had previously been cultured in MRS growth medium, harvested in the exponential phase, centrifuged and dispersed in water, prior to being added to the flour. Flour fermentation was performed at 30°C in a proofing cabinet at 70% relative humidity (RH) for about 18 hours.

Soaking in lactic acid

The rye flour used in the crisp bread was soaked in lactic acid according to the following description: 1000g flour was mixed with 1000g 0.15 M lactic acid at

30°C.

Soaking in acidic phosphate buffer

The rye flour used in the rye crisp bread was soaked in phosphate buffer according to the following description: 1000g flour was mixed with 1000g 0.1 M phosphate buffer, pH 4.0, 30°C.

Doughs with the following composition were produced from the pre-treated or non-pre-treated flours:

1. No pre-treatment

345g rye flour + 300g water + 7g NaCl was mixed for 4 min in a Hobardt mixer equipped with a dough hook.

2. Fermentation - lactic acid bacteria added

450g fermented flour/water mixture (bacteria added) + 150g untreated rye flour + 7g NaCl was mixed for 4 min in a Hobardt mixer equipped with a dough hook.

3. Lactic acid

450g flour/water mixture in lactic acid + 150g untreated rye flour + 7g NaCl was mixed for 4 min in a Hobardt mixture equipped with a dough hook.

4. Phosphate buffer

450g flour/water mixture in phosphate buffer + 150g untreated rye flour + 7g NaCl was mixed for 4 min in a Hobardt mixer equipped with a dough hook..

The proportion of pre-treated flour in doughs 2 to 4 was thus 60%.

About 100g of dough was rolled to 0.5mm thickness and baked at 240°C for 10 min.

Analysis of acrylamide was effected by Norsk Matanalyse

AS, Oslo, Norway.

Results

The results are presented in Table 1 below.

Pre-treatment of rye flour	pH after pre-treatment	Acid equivalent ml 0.1N NaOH/10g	Acrylamide µg/kg product (ppb)	Acrylamide % reduction*
No-pretreatment	6.00	4.0	3500	-
Fermentation - lactic acid bacteria added	3.70	18	890	75
Soaked in lactic acid	4.05	20	1000	71
Soaked in phosphate buffer	4.74	11	1400	60

*Reduction relative to crisp bread with no pre-treatment.

The pH in the rye flour/water mixture with bacteria added decreased during the pre-treatment. This shows that fermentation had occurred with corresponding acid production. The pH of the mixture with lactic acid and phosphate buffer added was as expected. At the applied conditions, adding lactic acid was effective in lowering pH and was thus similar to the controlled fermentation. The acid equivalent values reflected the changes seen in pH, with higher levels corresponding to the most extensive changes in pH.

Acrylamide levels in crisp bread reflected the various pre-treatments tested. With no pre-treatment, the acrylamide level was high. The use of 60% rye flour fermented by the adding of lactic acid bacteria caused a 75% reduction in acrylamide compared with crisp bread

produced without pre-treatment. The reduction was 71% when 60% of the flour was soaked in lactic acid as a pre-treatment. With phosphate buffer, the reduction in acrylamide was 60%.

It is likely that the pre-treatment can be optimised in several ways. First, the present experiments were performed with pre-treatment of only 60% of the total flour in the crisp bread. Doughs based on 100%-fermented flour may be used. Alternatively, the fermented flour may be fully or partly dried prior to application in the crisp bread dough, thus making it possible to adjust the viscosity of the dough to a level appropriate for rolling. Combinations of fermentation and acids may be effective in further lowering acrylamide levels, as well as sole adding of lactic or other acids to higher levels, and/or lower pH values.

Example 2

Potato Products (French fries and potato "crisps" (American "chips"))

Ingredients

Potatoes of the varieties Saturna and Beate were obtained from Department of Horticulture and Crop Sciences, Agricultural University of Norway, Ås. The potatoes were stored at 8°C from harvest until three weeks prior to processing when storage temperature was reduced to 4°C.

Palm oil was obtained from Denofa AS, Frederikstad, Norway. The oil had maximum 0.05% free fatty acids, an iodine number of 60, a peroxide value of 0.5 mekv/kg and an anisidin number of 5.0. Fatty acid composition was: 12% linoleic acid, 42% oleic acid and 45% saturated fatty acids.

Citric-phosphate buffer, pH 4, was prepared by mixing 1.8 L 0.1 M citric acid and 1.2 L 0.2 M di-sodium-phosphate.

The *Lactobacillus* strain NCIMB 40450 was used. Bacteria cells were grown and harvested in the logarithmic growth phase by centrifugation and resuspended in 1% salt brine.

The soaking solutions used were:

Brine for fermentation:	1% NaCl with the addition of bacteria until 1x 10^6 cells/ml
Brine with lactic acid:	1% NaCl and 1% lactic acid in water
Brine with buffer:	1% NaCl in citrate/phosphate buffer, pH4

Pre-treatment of potatoes

Potatoes (var. Saturna) were peeled and sliced (Robot Vertical Cutter 2, Robot Coupe SA, Le Perreux, France) to 1.5 mm thickness. Potatoes (var. Beate) were peeled and cut with a knife into 6 x 6-mm sticks. The slices and sticks (200g) were immediately added to 400 mL of one of the brines. Samples with acids were left at room temperature for 5 hrs while fermentations were allowed to proceed in an incubator at 30°C for 5 hrs. Control samples were rinsed in water and deep-fried without delay.

Deep frying

The potatoes were dried with paper towels and deep-fried as 150-g portions in palm oil at 170°C in a Nuovo Elframo, Model EB (Bergamo, Italy) fryer. For potato crisps the frying time was 3 min and for French fries 8 min.

Analyses

Dry matter was determined in a vacuum oven at 70°C over-night. pH of brines was determined using a pH-meter. Soluble solids of potatoes were determined as °Brix using a Metler Toledo RE40 refractometer. Samples were homogenised and a few drops of the homogenates were applied on the refractometer. °Brix is given as g sucrose/100g sample.

Accredited analyses of acrylamide were carried out at Steins Laboratorium, Denmark.

Results

The °Brix-value of the potatoes for the crisps production (var. Saturna) was 6.1, and of the potatoes for the production of French fries (var. Beate) 6.8.

Results from the analyses of deep-fried products are shown in Table 2 below.

Table 2

		Dry matter		Acrylamide	
Pre-treatment	pH in brine after pre-treatment	g/100g fried product	µg/kg dry matter	µg/kg product	% reduction (product)
Potato crisps					
-control	6.7*	95.7	951	910	0
-fermented	4.6	95.8	397	380	58
-lactic acid	5.6	95.1	725	690	24
-buffer	4.2	95.1	673	640	30
French fries					
-control	6.7*	86.0	744	640	0
-fermented	4.3	85.3	469	400	38

* tap water

All samples within each of the product groups were deep-fried for the same period of time, rather than being deep-fried until a certain product colour. Any difference in acrylamide levels thus reflects the ability of each treatment to prevent the formation of acrylamide, regardless of the colour that might be formed during deepfat-frying.

Other plant materials, e.g. carrots and other vegetables, may be processed similarly and show significant reductions in acrylamide content. Thus for example carrot crisps (made using 1.5mm slices of carrot) show a 93% reduction in acrylamide content following fermentation as compared with a water rinse in place of fermentation.

Claims:

1. The use of a lactic acid producing microorganism for the treatment of a food material to reduce acrylamide production in subsequent cooking thereof.

2. Use as claimed in claim 1 of a lactic acid bacterium.

3. The use of a physiologically acceptable acid for the treatment of a food material to reduce acrylamide production in subsequent cooking thereof.

4. Use as claimed in claim 3 of an acid selected from lactic, citric, phosphoric and hydrochloric acids.

5. A process for the preparation of ready to cook french fried potatoes which process comprises chopping potatoes, fermenting the chopped potatoes with a lactic acid producing microorganism, frying the fermented chopped potatoes, and optionally loading the fried fermented chopped potatoes into a container.

6. A process for the preparation of ready to cook french fried potatoes which process comprises chopping potatoes, treating the chopped potatoes with a physiologically acceptable acid, frying the acid treated potatoes, and optionally loading the fried potatoes into a container.

7. A container, preferably a sealed container, containing oven-ready french fried potatoes produced by frying chopped potatoes pre treated with a lactic acid producing microorganism and/or a physiologically acceptable acid.

8. A process for the production of a food product

which comprises fermenting a granulated or crushed carbohydrate-containing plant material with a lactic acid producing microorganism, optionally formulating the fermented material into a shaped product, and cooking to produce said food product.

9. A process for the production of a food product which comprises treating a granulated or crushed carbohydrate-containing plant material with a physiologically tolerable aqueous acid, optionally formulating the treated material into a shaped product, and cooking to produce said food product.

10. A process as claimed in either of claims 8 and 9 further comprising packaging the food product into sealed sterilized containers.

11. Food products produced by a process according to any one of claims 5, 6, 8, 9 and 10 or according to a use as claimed in any one of claims 1 to 4.